A RESUMÉ OF RECENT TECHNICAL METHODS FOR THE NERVOUS SYSTEM.

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GOLGI'S METHODS.[1]—1. *Silver Process.*

Small pieces (one to two cubic centimetres) are placed in a two-per-cent solution of potassium bichromate (or Müller's fluid) containing camphor. The concentration is increased at each renewal of the fluid up to five per cent. The length of time of the hardening varies according to the amount of the material, the concentration of the solution, and the temperature. A constant temperature (20–25° Cent.) is advantageous. If the thermostat is not used, in summer, portions of the brain are hardened fifteen to twenty, rarely forty to fifty days, and in winter five to six, to sixteen weeks. To avoid all post-mortem changes, the author injects a two-per-cent solution of bichromate, containing five to six per cent of gelatin, into the carotids. The hardening process may be hastened by replacing a part of the hardening fluid by Erlicki's fluid in increasing proportions. A still more rapid procedure consists in the after-hardening in a mixture of eight parts of a two-and-one-half-per-cent solution of bichromate and two parts of a one-per-cent solution of osmic acid.

The hardened pieces are placed in a large volume of a three-fourths of one-per-cent solution of nitrate of silver,

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[1] C. Golgi, "Recherches sur l'histologie des centres nerveux." Archives italiennes de Biologie, Vols. iii., iv., 1883.

C. Golgi, "Sulla fina anatomia degli organi centrali del sistema nervoso." Review in Neurolog. Centralblatt, No. 13, 1886.

which must be renewed at intervals until the chromate of silver precipitate no longer forms. If the hardening does not seem complete, a one-half-per-cent silver solution should be used. If the hardening has been protracted, the silver solution is increased to a strength of one per cent. The blocks remain in the silver solution twenty-four to thirty hours; a longer immersion is not usually injurious. After cutting, the sections are carefully washed in alcohol, cleared first in creasote and then in turpentine. If the sections are carefully washed, they need not be protected from light. During the hardening, it is advisable to test portions occasionally with the silver solution, for the method is somewhat uncertain.

2'. *The Sublimate Process.*—The same results may be obtained if the hardened pieces are immersed in a one-half-per-cent-solution of bichloride of mercury instead of the silver solution. The immersion in the sublimate solution requires several weeks. This method has the advantage that very large masses, even the entire brain, may be stained *in toto*, thus facilitating the preparation of serial sections.

The results accomplished by Golgi's methods have recently been highly commended by Forel.[1] The astonishing selective affinity that these metallic combinations exhibit for the ganglion-cell with all of its processes, and for the intricate network derived from the axis-cylinder process of certain ganglion-cells, and for the transitional area from the ganglion-cell to the nerve-fibre opens new histological and pathological territories. Perhaps some of the incomplete chapters in cortical pathology will receive contributions from this method, and a re-examination by this process is required of cases in which delicate changes in the ganglion-cells are suspected, but which have eluded the previous methods.

A great advantage of Golgi's process is the contrast afforded between the colored and the uncolored elements, as the ganglion-cells and their immediate adnexa are sharply

[1] Prof. A. Forel, "Einige hirnanatomische Betrachtungen und Ergebnisse." Archiv f. Psychiatrie, Band xviii., No. 1.

delineated in black against a faint yellowish back-ground. This black color in the silver process is probably due to a precipitate of chromate of silver; in the sublimate method, to one of the oxides of mercury. This contrast of color renders thick sections rather more useful than thin ones, for in the latter many of the processes of an individual cell may be lost. A peculiar feature of this method is its property of staining only here and there scattered and isolated elements in the section—a less confusing picture than if the whole section were uniformly stained.

Bleuler[1] has succeeded with the stain and has obtained fac-similes of many of Golgi's plates. A resumé of Golgi's remarkable propositions concerning the ganglion-cells, many of which are sustained by Forel, may be found in Forel's[2] paper. Pal,[3] in discussing the results obtained by a modification of Golgi's methods, observes that the neuroglia-cells stain in the same way as the ganglion-cells, and that the neuroglia-cells of the white substance react the best. Pal obtained the best results in the cortex; the cerebellar sections were less successful, and the results in the cord were almost entirely unsuccessful. The one disadvantage of the method is its uncertain results.

Pal's Modification of Golgi's Methods.—1. If the sections prepared by the sublimate process are inspected with the naked eye, they contain whitish, opaque spots. The sections are immersed in a one-half to one-fourth-per-cent solution of sodium sulphide until the spots become blackened. The sections are then washed and may subsequently be advantageously stained with magdala-red.

2. The silver preparations may be treated in the same manner. The sections are placed in the sulphide of sodium solution, which gives a black sulphide of silver precipitate in the ganglion-cells. The author claims that this precipitate is more stable than that of chromate of silver occurring in Golgi's original process. These prepara-

[1] Correspondenzbl. f. Schweizer Aerzte, March 15th, 1886.

[2] L. c.

[3] J. Pal, "Ein Beitrag zur Nervenfärbentechnik;" Medicin. Jahrbücher der K. K. Gesellschaft, 1886, No. 9.

tions do not show the network of the processes as well as the modified sublimate method.

PAL'S MODIFICATION OF WEIGERT'S METHOD.—Pal proposes a modification of Weigert's method.[1] The copper immersion is dispensed with, and two to three cubic centimetres of lithium carbonate are added to Weigert's hæmatoxylin solution. Sections are stained twenty-four to forty-eight hours. The sections are washed in water; if the sections are not stained a deep blue color, one to two cubic centimetres of lithium carbonate are added to the water used for washing. The sections are then transferred to a one-fourth-per-cent solution of permanganate of potassium for twenty to thirty seconds, where they present the same appearances as in decolorizing with the ordinary method. The specimens are then transferred to the following solution:

Oxalic Acid	1
Potassium Sulphite	1
Aq. Dest	200

for a few seconds. The sections are better adapted for subsequent contrast staining than those obtained by the usual method. If flocculi persist on the surface of the sections while in the oxalic acid mixture, the sections are again rapidly passed through the permanganate solution, and then replaced in the oxalic acid mixture.

Sections prepared by Golgi's methods may be stained by this modified Weigert's method, by placing them for twenty-four hours in a five-per-cent solution of chromic acid previous to staining them in the hæmatoxylin solution. Pal also recommends a one in four hundred aqueous solution of potassium permanganate, for removing the color from all structures except the medullated nerve fibres, in

[1] Freud (Vienna), reviewing this modification for the Neurolog. Centralblatt, March, 1887, recommends it, and observes that the differentiation is sharper than that obtained by the ordinary method. Serial sections between two adherent films of celloidin should be first placed for a few minutes in the prussiate-borax solution of Weigert, and then be subjected to the differentiating solutions of Pal.

sections of the nervous system hardened in one-per-cent osmic acid.

ALKANNA FOR MYELIN STAINING.—Achard[1] recommends a concentrated alcoholic solution of the cortical portions of alkanna root for the central nervous system and also for the peripheral nerves. Fragments of the roots are macerated in a relatively large volume of ninety-per-cent alcohol for several days in a well-corked flask, until the solution has a garnet-red tint. The solution must frequently be prepared anew. Specimens hardened by the chrome salts are left in this solution about two hours, avoiding the access of air by using carefully closed dishes. If the hardening has been protracted, the sections should not remain too long in the staining reagent, for the color may become diffuse. The stained sections are rapidly washed in water and mounted in glycerin. The myelin has a deep brown color, and the result is similar to that given by Weigert's method, with the disadvantage, however, that the sections cannot be mounted in balsam, for this procedure removes the color completely. The sections may be stained with carmine first and subsequently with the alkanna.

CHANGES PRODUCED IN GANGLION CELLS BY HARDENING REAGENTS.—Trzebinski[2] has studied the changes produced in the ganglion cells of the spinal cord in dogs and rabbits, by using the following reagents:

1. Müller's fluid (five to six weeks), followed by alcohol.
2. Alcohol ninety-six-per-cent.
3. Chromic acid, followed by Müller's fluid or alcohol.
4. Hardening in a ten-per-cent solution of bichloride of mercury for eight days and subsequent hardening in alcohol containing five-per-cent of iodine. Reagents 1 and 3 produce very serious alterations in the contour and finer

[1] Ch. Achard, "Sur l'emploi de la Teinture d'orcanette dans la technique histologique;" Archives de Physiologie, 1887, No. 2.

[2] Dr. S. Trzebinski, "Einiges über die Einwirkung der Härtungsmethoden auf die Beschaffenheit der Ganglienzellen im Rückenmark der Kaninchen und Hunde." Virchow's Archiv, Band 107, No. 1.

structure of the cell, which have undoubtedly been ascribed by some authors to pathological processes.

In specimens hardened by reagent 4, these changes are reduced to a minimum. Vacuoles and pale or faintly stained ganglion cells [1] are absent. The pericellular spaces are small and the finer structural details resemble closely the appearances seen in the cells when examined in the fresh condition. The condition of the cells after alcoholic hardening closely resembles the result obtained by the sublimate hardening, and the author indicates a preference for the latter method.

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[1] Dr. F. Kreyssig, Virchow's Archiv, Band 102; H. Koneff, Fortschrit. d. Med., 1886, No. 23.